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(81) Designated States (*national*): AE, AG, AL, AM, AT, AU,  
AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU,  
CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE,  
GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR,  
KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK,  
MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT,  
RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR,  
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GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW),  
Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM),  
European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE,  
ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE,  
SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA,  
GN, GQ, GW, ML, MR, NE, SN, TD, TG).

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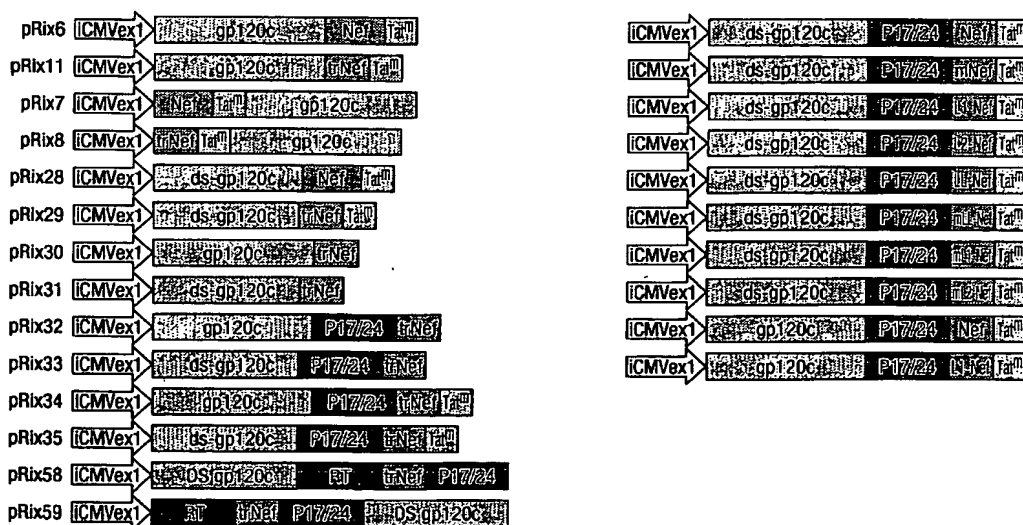
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[Continued on next page]

(54) Title: VACCINE

A Schematic representation of further constructs



(57) Abstract: The invention relates to polynucleotides for DNA vaccination which polynucleotides encode an HIV envelope protein or fragment or immunogenic derivative fused to an additional HIV protein selected from a non-structural protein or capsid protein or fragment or immunogenic derivative thereof. Preferably the HIV envelope molecule is gp120 and preferred fusions include one or more of HIV Nef, Gag, RT or Tat. Preferably the HIV envelope molecule is non-glycosylated in mammalian cells.

**WO 2004/041851 A3**



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*For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.*

# INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/12429

## A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C07K14/16 A61K39/21 C12N15/48 A61K48/00 C12N15/62

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C07K C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the International search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, PAJ, BIOSIS, EMBASE, MEDLINE, CHEM ABS Data, Sequence Search

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WOODBERRY T ET AL: "Immunogenicity of a human immunodeficiency virus (HIV) polytope vaccine containing multiple HLA A2 HIV CD8+ cytotoxic T-cell epitopes" JOURNAL OF VIROLOGY, THE AMERICAN SOCIETY FOR MICROBIOLOGY, US, vol. 73, no. 7, July 1999 (1999-07), pages 5320-5325, XP002162348 ISSN: 0022-538X	1-6, 10-12, 17-31
Y	the whole document ----- -/--	13-16

☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

### \* Special categories of cited documents:

\*A\* document defining the general state of the art which is not considered to be of particular relevance

\*E\* earlier document but published on or after the International filing date

\*L\* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

\*O\* document referring to an oral disclosure, use, exhibition or other means

\*P\* document published prior to the International filing date but later than the priority date claimed

\*T\* later document published after the International filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

\*X\* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

\*Y\* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

\*8\* document member of the same patent family

Date of the actual completion of the international search

31 August 2004

Date of mailing of the International search report

11.01.05

Name and mailing address of the ISA

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# INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/12429

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	LIU W J ET AL: "Papillomavirus Virus-like Particles for the Delivery of Multiple Cytotoxic T Cell Epitopes" VIROLOGY, ACADEMIC PRESS, ORLANDO, US, vol. 273, no. 2, 1 August 2000 (2000-08-01), pages 374-382, XP004436241 ISSN: 0042-6822	1-3,6, 11,12, 17-21, 23-31
Y	the whole document	4,5, 13-16,22
X	WO 01/27291 A (LANGLADE DEMOYEN PIERRE ; FIRAT HUESEYIN (FR); LEMONNIER FRANCOIS ( ) 19 April 2001 (2001-04-19)	1-6, 10-12, 17-31
Y	figure 5; example 11; tables 5-8	13-16
X	WO 02/32943 A (CHADRABARTI BIMAL K ;HUANG YUE (US); US GOVERNMENT (US); NABEL GAR) 25 April 2002 (2002-04-25)	1-3,6, 11,13, 14, 17-21, 23-31
Y	the whole document	4,5,12, 15,16,22
X	EP 0 577 894 A (GREEN CROSS KOREA) 12 January 1994 (1994-01-12)	1-3, 10-12, 17-21, 23-31
Y	the whole document	4,5,13, 15,16,22
X	BENKO D M ET AL: "A NOVEL HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 PROTEIN TEV SHARES SEQUENCES WITH TAT ENV AND REV PROTEINS" JOURNAL OF VIROLOGY, vol. 64, no. 6, 1990, pages 2505-2518, XP002285841 ISSN: 0022-538X	1-3, 11-13, 17-21, 23-31
	the whole document	
X	SALFELD J ET AL: "A TRIPARTITE HIV-1 TAT-ENV-REV FUSION PROTEIN" EMBO JOURNAL, OXFORD UNIVERSITY PRESS, SURREY, GB, vol. 9, no. 3, 1 March 1990 (1990-03-01), pages 965-970, XP000113784 ISSN: 0261-4189	1-3, 11-13, 17-21, 23-31
	the whole document	
Y	WO 98/41536 A (DESROSIERS RONALD C ; HARVARD COLLEGE (US); REITTER JULIE N (US)) 24 September 1998 (1998-09-24)	11
	the whole document	
	-/--	

## INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/12429

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT		
Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	DOE B ET AL: "INDUCTION OF HIV-1 ENVELOPE (GP120)-SPECIFIC CYTOTOXIC T LYMPHOCYTE RESPONSES IN MICE BY RECOMBINANT CHO CELL-DERIVED GP120 IS ENHANCED BY ENZYMATIC REMOVAL OF N-LINKED GLYCANS" EUROPEAN JOURNAL OF IMMUNOLOGY, WEINHEIM, DE, vol. 24, no. 10, 1994, pages 2369-2376, XP000672290 ISSN: 0014-2980 the whole document	11
Y	BOTARELLI P ET AL: "N-GLYCOSYLATION OF HIV-GP120 MAY CONSTRAIN RECOGNITION BY T LYMPHOCYTES" JOURNAL OF IMMUNOLOGY, THE WILLIAMS AND WILKINS CO. BALTIMORE, US, vol. 147, no. 9, 1 November 1991 (1991-11-01), pages 3128-3132, XP000673664 ISSN: 0022-1767 the whole document	11,12,14
Y	STEFANIE A ET AL: "Increased Immune Response Elicited by DNA Vaccination with a Synthetic gp120 Sequence with Optimized Codon Usage" JOURNAL OF VIROLOGY, THE AMERICAN SOCIETY FOR MICROBIOLOGY, US, vol. 72, no. 2, February 1998 (1998-02), pages 1497-1503, XP002278034 ISSN: 0022-538X the whole document	13,14
Y	KOTSOPLOULOU E ET AL: "A Rev-independent human immunodeficiency virus type 1 (HIV-1)-based vector that exploits a codon-optimized HIV-1 gag-pol gene" JOURNAL OF VIROLOGY, THE AMERICAN SOCIETY FOR MICROBIOLOGY, US, vol. 74, no. 10, May 2000 (2000-05), pages 4839-4852, XP002140792 ISSN: 0022-538X the whole document	13,14
Y	WO 02/36792 A (CATCHPOLE IAN RICHARD ;RHODES JOHN RICHARD (GB); GLAXO GROUP LTD ) 10 May 2002 (2002-05-10) cited in the application the whole document	15,16
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# INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/12429

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	IGLESIAS, ENRIQUE ET AL: "Chimeric proteins containing HIV-1 T cell epitopes: Expression in E. coli, purification and induction of antibodies in mice" JOURNAL OF BIOCHEMISTRY, MOLECULAR BIOLOGY AND BIOPHYSICS , 5(2), 109-122 CODEN: JBMBF6; ISSN: 1025-8140, 2001, XP009035847 the whole document	1-6, 10-31
X	CA 2 430 702 A (CT DE INGENIERIA GENETICA Y BI) 6 September 2002 (2002-09-06) the whole document	1-6, 10-31
E	WO 2004/041852 A (GLAXO GROUP LTD ; ERTL PETER FRANZ (GB)) 21 May 2004 (2004-05-21) the whole document	1-6, 10-31

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/EP 03/12429

## Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:  

Although claim 28 is directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.
2. ☐ Claims Nos.:  
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

## Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☒ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:  

1-6, 10-31
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☒ No protest accompanied the payment of additional search fees.

## FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 6 (completely) 1-3, 10-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter.

1.1. claims: 6 (completely) 1-3, 11-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter, wherein the at least one structural or capsid protein or fragment or immunogenic derivative thereof is selected from Nef, and subject-matter related thereto.

1.2. claims: 1-3, 10-13, 15-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter, wherein the at least one structural or capsid protein or fragment or immunogenic derivative thereof is selected from Gag, and subject-matter related thereto.

1.3. claims: 1-3, 11-13, 15-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter, wherein the at least one structural or capsid protein or fragment or immunogenic derivative thereof is selected from RT, and subject-matter related thereto.

1.4. claims: 1-3, 11-13, 15-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter, wherein the at least one structural or capsid protein or fragment or immunogenic derivative thereof is selected from Tat, and subject-matter related thereto.

## FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

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2. claims: 4, 5, 22 (all completely), 1-3, 10-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter, wherein the polynucleotide encodes a gp120, RT, Gag and Nef-containing fusion protein, and subject-matter related thereto.

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3. claims: 7,8 (all completely), 1-3, 11-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter, wherein the polynucleotide encodes a gp120, Tat and Nef-containing fusion protein, and subject-matter related thereto.

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4. claims: 9 and 32-34 (all completely), 1-3, 10-21, 23-31 (all partially)

A polynucleotide which comprises a sequence encoding an HIV envelope protein or fragment or immunogenic derivative thereof, fused to at least one sequence encoding an HIV non-structural or capsid protein or fragment or immunogenic derivative thereof, operably linked to a heterologous promoter, wherein the polynucleotide encodes a gp120-Gag-Nef-Tat fusion, and subject-matter related thereto. A polynucleotide encoding an HIV Tat molecule or fragment or immunogenic derivative in a fusion with at least two further HIV antigens, and subject-matter related thereto.

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# INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/EP 03/12429

Patent document cited in search report		Publication date	Patent family member(s)	Publication date
WO 0127291	A	19-04-2001	AU 1023401 A WO 0127291 A1 US 2002164721 A1	23-04-2001 19-04-2001 07-11-2002
WO 0232943	A	25-04-2002	AU 8499901 A CA 2419822 A1 EP 1311686 A2 WO 0232943 A2 US 2004033487 A1	29-04-2002 25-04-2002 21-05-2003 25-04-2002 19-02-2004
EP 0577894	A	12-01-1994	KR 172970 B1 DE 69223952 D1 DE 69223952 T2 EP 0577894 A1 JP 6321997 A US 5580773 A US 5443828 A	01-02-1999 12-02-1998 13-08-1998 12-01-1994 22-11-1994 03-12-1996 22-08-1995
WO 9841536	A	24-09-1998	AU 6535898 A WO 9841536 A1	12-10-1998 24-09-1998
WO 0236792	A	10-05-2002	AU 1250502 A CA 2427952 A1 EP 1334201 A2 WO 0236792 A2 JP 2004512837 T US 2004082531 A1	15-05-2002 10-05-2002 13-08-2003 10-05-2002 30-04-2004 29-04-2004
CA 2430702	A	06-09-2002	BR 0206823 A CA 2430702 A1 WO 02068654 A2 JP 2004518443 T US 2004073008 A1 EP 1371730 A2	25-02-2004 06-09-2002 06-09-2002 24-06-2004 15-04-2004 17-12-2003
WO 2004041852	A	21-05-2004	WO 2004041852 A2	21-05-2004

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(71) Applicant (for all designated States except US): **GLAXO GROUP LIMITED** [GB/GB]; Glaxo Wellcome House, Berkeley Avenue, Greenford, Middlesex UB6 0NN (GB).

(72) Inventor; and

(75) Inventor/Applicant (for US only): **ERTL, Peter, Franz** [GB/GB]; GlaxoSmithKline, Gunnels Wood Road, Stevenage, Hertfordshire SG1 2NY (GB).

(74) Agent: **PRIVETT, Kathryn, Louise**; GlaxoSmithKline (CN925.1), 980 Great West Road, Brentford, Middlesex TW8 9GS (GB).

(81) Designated States (*national*): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) Designated States (*regional*): ARIPO patent (BW, GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

**Published:**

— without international search report and to be republished upon receipt of that report

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: VACCINE

(57) Abstract: The invention relates to polynucleotides for DNA vaccination which polynucleotides encode an HIV envelope protein or fragment or immunogenic derivative fused to an additional HIV protein selected from a non-structural protein or capsid protein or fragment or immunogenic derivative thereof. Preferably the HIV envelope molecule is gp120 and preferred fusions include one or more of HIV Nef, Gag, RT or Tat. Preferably the HIV envelope molecule is non-glycosylated in mammalian cells.



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